

Age and Growth of *Glossocardia obesa*, a “Large” Bivalve in a Submarine Cave Within a Coral Reef, as Revealed by Oxygen Isotope Analysis

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Abstract. Dark submarine caves are unique habitats from which most coastal species are excluded because of darkness, marked oligotrophy, and reduced water circulation. Many bivalves have been found in caves of tropical West Pacific coral reefs, but nearly all reported species are very small in adult shell size, generally <5 mm in length. *Glossocardia obesa* (Reeve, 1843) is an exceptionally large bivalve in the cave fauna and grows to 80 mm in adult shell length. Since live specimens of *G. obesa* are apparently very uncommon, there has been no study on its life-history traits. We performed an oxygen isotope analysis of a *G. obesa* (72 mm in shell length) collected alive in a submarine cave at Okinawa Islands, Japan. The $\delta^{18}\text{O}$ record is divided into early- and late-growth phases. There is no systematic change in $\delta^{18}\text{O}$ value in the early-growth phase, whereas the $\delta^{18}\text{O}$ record for the late-growth phase contains six cycles. Comparing the $\delta^{18}\text{O}$ -derived temperatures from *G. obesa* with water-temperature records and $\delta^{18}\text{O}$ value of seawater in the cave shows that the $\delta^{18}\text{O}$ cycles of *G. obesa* reflect seasonal variation in water temperature. This result demonstrates that the oxygen isotope analysis can usefully be employed to estimate growth and age in this species.

INTRODUCTION

Sheltered, submarine caves developed within coral reefs are inhabited by unique invertebrate communities that include “living fossil” species and relatives of deep-sea taxa (Jackson et al., 1971; Jackson & Winston, 1982; Vermeij, 1987; Harmelin, 1997). Many workers have addressed the ecological and evolutionary significance of the cryptic communities (Reitner & Gautret, 1996;

Kano et al., 2002; Motchurova-Dekova et al., 2002; Kano & Kase, 2008; Ubukata et al., 2009). Kase & Hayami (1992) and Hayami & Kase (1993, 1996) investigated cavernicolous bivalves from many caves in Ryukyu Islands, Bonin Islands, Philippine Islands, Saipan, Palau, and Guam, and reported >60 species, many of which share the following characteristics: (1) very small adult size (usually <5 mm in length); (2) unusually large prodissoconch I and an absence of

prodissoconch II in many species, implying nonplanktotrophic development; and (3) persistent denticles on the provinculum retained until the adult stage in many pteriomorph species, suggesting significant paedomorphosis by progenesis. Kase & Hayami (1992) interpreted these characteristics to indicate a relatively small number of larvae and a predominantly K-selected reproductive strategy. The authors regarded these ecological features as adaptations to nutritional deficiency in the submarine caves, as widely documented by Fichez (1990, 1991).

Although most of the bivalves in the caves are very small in adult shell size, the trapeziid *Glossocardia obesa*, a shallow infaunal suspension feeder, is an exceptionally large bivalve in the cave fauna (Hayami & Kase, 1993). Shell length exceeds 80 mm and the shell volume is over $\times 2000$ greater than those of other cavernicolous microbivalves. Hayami & Kase (1992, 1996) reported that the soft part of *G. obesa* is much smaller than that expected for their shell size; however, their soft bodies are still much larger than those of other cavernicolous microbivalves.

This species, distributed in the tropical West Pacific region, lives in both the open sea and cryptic environments such as submarine caves developed within coral reefs (Matsukuma & Habe, 1995). Morton (1979) described this bivalve as a deepwater species, and it has also been recorded in shelly gravel bottoms at between 5- and 60-m water depth (Okutani, 2000). However, there has been no study of the life-history traits of *G. obesa* because it is very rare to encounter living individuals in either the open sea or cryptic environments such as submarine caves.

In this paper, we examined the pattern and rate of shell growth of a single specimen of *G. obesa* collected alive in a submarine cave developed within a coral reef (Figure 1), from the perspective of oxygen isotope profiles. This method is commonly used to decipher the life-history traits of living and extinct bivalve species (e.g., Jones et al., 1983, 1986; Tanabe & Oba, 1988; Goodwin et al., 2001; Schöne et al., 2003; Watanabe et al., 2004). The proportion of different isotopes of oxygen present in the shell carbonate reflects the ambient water temperature in which the shell was deposited because in colder water more of the heavier ^{18}O isotope is precipitated into the shell, while at higher temperatures relatively more of the lighter ^{16}O isotope is precipitated.

SHELL DESCRIPTION AND STUDY AREA

We obtained a shell of *G. obesa* collected alive by a scuba diver in the submarine Daidokutsu cave located in the northeastern coast of Ie Island, Okinawa Island (Figure 1), in early August 2004. The cave's entrance lies about 19 m below sea level. The cave is 40 m long,

dark inside, and deepens inward to its deepest point at 29 m below sea level. The floor is covered by >1.4 m of calcareous mud (Kitamura et al., 2007a). So-called living fossils, including the gastropod *Neritopsis radula* and the bivalve *Pycnodonte taniguchii*, have been found alive in the cave (Hayami & Kase, 1992; Kase & Hayami, 1992). In addition, at least 36 species of cavernicolous microbivalves have been identified from the sediments of the cave (Kitamura et al., 2007a). Kitamura et al. (2007b) measured water temperatures hourly in the cave from July 26, 2003 to July 6, 2004, documenting seasonal variation from 21°C (February) to 29°C (late August to early September; Figure 2A).

METHODS

Water samples for oxygen isotope analyses were collected within Daidokutsu and upon the reef slope at 30-m depth on July 2, 2007 (10:50–11:15 a.m. Japan time; Figure 1). Oxygen isotope analyses of the water ($\delta^{18}\text{O}_w$) were undertaken at the Geo-Science Laboratory in Nagoya, Japan, using a Finnigan MAT delta S. $\delta^{18}\text{O}_w$ ratios are reported relative to Vienna Standard Mean Ocean Water (V-SMOW), and the analytical precision (1 SD) was better than $\pm 0.1\%$. Salinity and temperature were measured using a temperature–salinity meter at the time of collecting water samples.

The specimen of *G. obesa* was embedded directly in polyester resin without any chemical treatment, and cut along the axis of maximum growth. The section was ground with 1200 *sic* grit, polished with 3- μm Al_2O_3 powder, and photographed under transmitted light. Powdered carbonate samples were collected from the outer shell layer using an automated Micromill sampler at the Japan Agency for Marine-Earth Science and Technology (JAMSTEC), Japan (Sakai, 2007). The width of the sample grooves was 2 mm (80–120 mg in weight) from the umbo down to 52 mm, and the width was 1 mm (50–70 mg in weight) from the 52-mm point to the shell margin. The powdered carbonate samples received no additional thermal or chemical treatment prior to oxygen isotope analysis. The sample was analyzed using a mass spectrometer (IsoPrime, Micro-mass) at JAMSTEC. Individual samples were reacted with 100% phosphoric acid at 90°C. Oxygen isotope values of shell carbonate ($\delta^{18}\text{O}_c$) are reported relative to PeeDee belemnite, and the analytical precision (1 SD) was better than $\pm 0.1\%$. X-ray diffractometer analysis revealed that the shell of *G. obesa* consists entirely of aragonite.

RESULTS

There is no significant difference in both salinity and $\delta^{18}\text{O}_w$ between inside the cave and at 29-m depth (Table 1). The $\delta^{18}\text{O}_w$ –salinity relation is similar to the linear relationship proposed by Oba (1988) for surface

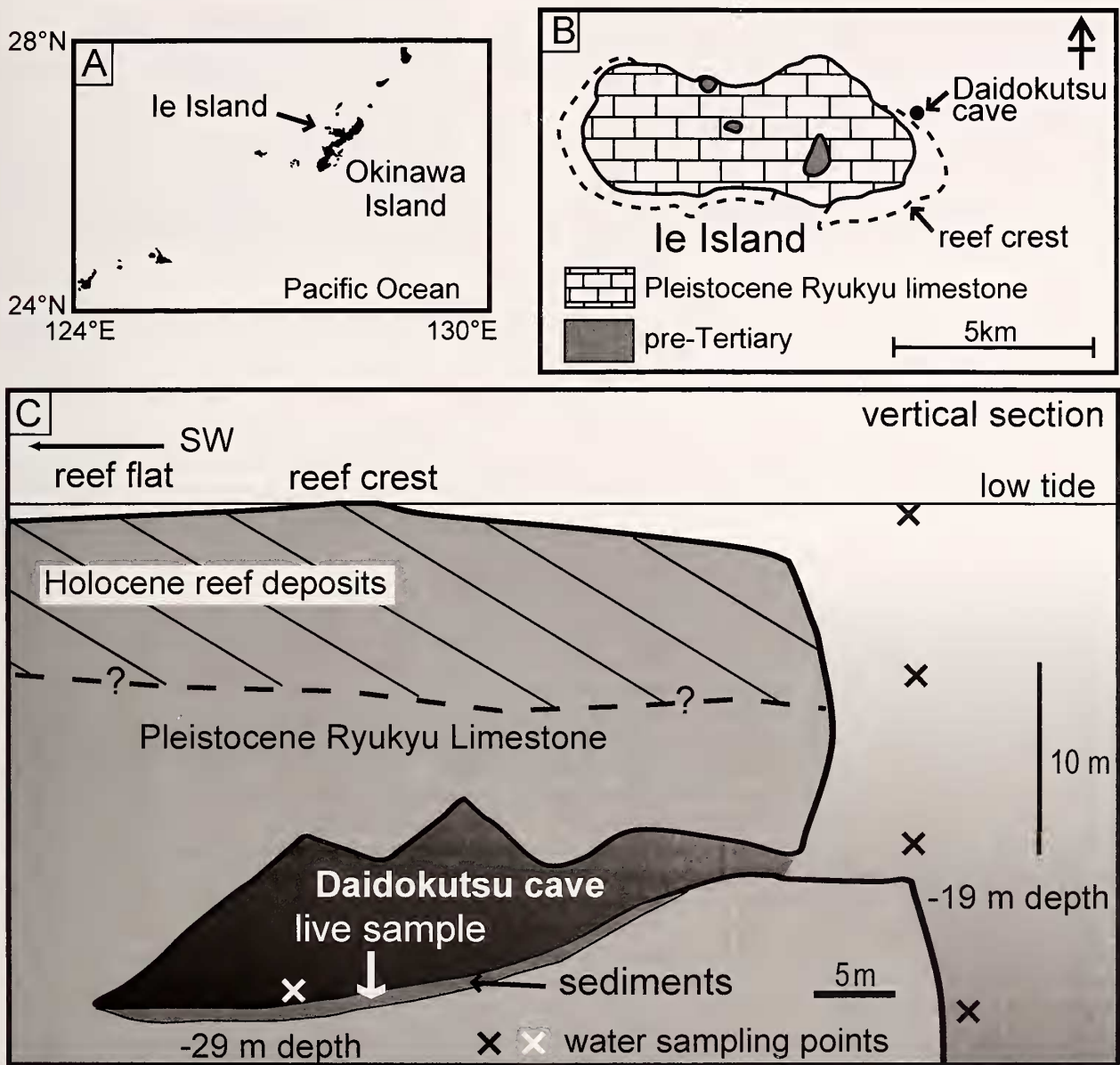


Figure 1. Locality map showing sampling site for *Glossocardia obesa* and Daidokutsu cave.

water extending from the East China Sea to the Kuroshio Current off the southern Japanese Islands, which is $\delta^{18}\text{O}_w = 0.203 S - 6.76$ ($S = \text{salinity}$).

Seven translucent growth lines with widths of 0.9–1.3 mm were observed in the outer shell layer under transmitted light (Figure 3). The $\delta^{18}\text{O}_c$ record is divided into early- and late-growth stages at a point located about 34 mm from the umbo (Figure 3). The early-growth stage shows no systematic variation in $\delta^{18}\text{O}_c$ values, which range from -1.8 to 0‰ . In contrast, the $\delta^{18}\text{O}_c$ record for the late-growth stage contains six cycles (Cycles 1 to 6; Figure 3) for which the wavelength decreases with distance from the umbo.

These six cycles are subdivided into four earlier cycles (1–4) and two later cycles (5 and 6). The $\delta^{18}\text{O}_c$ values in the former range from -2.6 to -0.8‰ , while in the latter from -2.0 to -1.2‰ . There are two translucent growth lines in Cycle 1, but only a single growth line was observed in each of the later cycles. These lines correspond to light oxygen isotope values in each cycle (Figure 3). There are anomalously high $\delta^{18}\text{O}_c$ values at a point located 26 mm from the umbo and at the shell margin. Such exceptionally heavy $\delta^{18}\text{O}_c$ values were often reported from other bivalves, such as *Mesodesma donacium* (Carré et al., 2005), but their origin remains poorly understood.

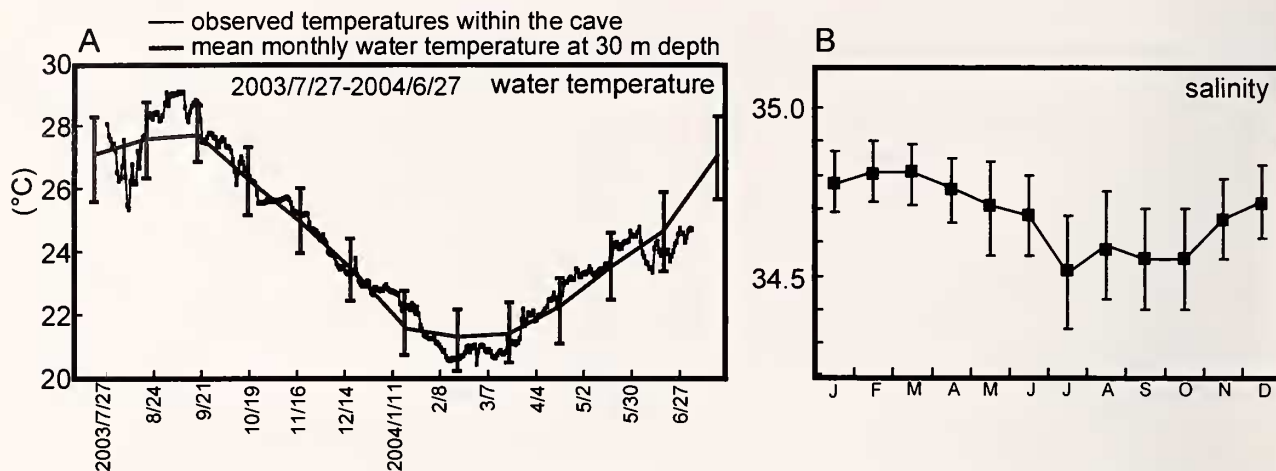


Figure 2. **A**, Comparison of observed temperatures with mean monthly water temperature at 29-m depth (the JODC). Data of observed temperatures within the cave are from Kitamura et al. (2007b); **B**, Seasonal variations in salinity at 29-m depth. Data are sourced from database compiled by the JODC. Error bars of monthly temperature and salinity represent standard deviation (1σ) from monthly value for the period between 1906 and 2003.

DISCUSSION

It is generally accepted that the oxygen isotopic composition of bivalves is influenced by the temperature and isotopic composition of the ambient seawater in which the animal formed its shell. As noted above, $\delta^{18}\text{O}_w$ in the cave is equal to that at 29-m water depth. In addition, the relationship between $\delta^{18}\text{O}_w$ and salinity inside and outside the cave is not considerably different than that previously proposed by Oba (1988). Therefore, seasonal variation in $\delta^{18}\text{O}_w$ within the cave can be estimated using salinity data at 30-m depth (S_{30m}), as compiled by the Japan Oceanographic Data Center (JODC; $1 \times 1^\circ$ grid cells, latitude $26\text{--}27^\circ\text{N}$ and longitude $127\text{--}128^\circ\text{E}$, data from the period 1874–2001), and the above $\delta^{18}\text{O}_w$ –salinity relationship (Oba, 1988).

Based on the data of JODC, S_{30m} ranges from 34.5 in summer to 34.8 in winter (Figure 2B). We regarded the $\delta^{18}\text{O}_w$ value in the cave for the entire year to be $0.3 \pm 0.03\text{‰}$ if the $\delta^{18}\text{O}_w$ –salinity relationship is adopted.

Table 1

Oxygen isotope values obtained for water sampled from within Daidokutsu cave and upon the reef slope off Ie Island, Okinawa, Japan. Locations of sampling points are shown in Figure 1.

Area	Reef slope				Daidokutsu
	0	10	20	30	
Depth (m)	0	10	20	30	29
Temperature ($^\circ\text{C}$)	29.2	28.0	27.5	26.6	25.2
Salinity	33.4	33.1	33.2	34.2	34.3
$\delta^{18}\text{O}_w$ (‰; vs. V-SMOW)				0.4	0.3

Water temperatures were calculated using the equation of Carré et al. (2005):

$$T(^{\circ}\text{C}) = (17.41 \pm 1.15) - (3.66 \pm 0.16)(\delta^{18}\text{O}_{\text{argonite}} - \delta^{18}\text{O}_w).$$

Except for two exceptionally heavy $\delta^{18}\text{O}$ values, all $\delta^{18}\text{O}_c$ -derived temperatures based upon using the equation of Carré et al. (2005) fell within the temperature range in the cave (Figure 4). The 6.5°C variation in $\delta^{18}\text{O}$ -derived temperature is ca. 81% of the annual range of temperatures in the cave from July 26, 2003 to July 6, 2004 (8°C). We therefore consider that the $\delta^{18}\text{O}_c$ cycles reflect seasonal fluctuations in water temperature. The relatively small amplitudes of Cycles 5 and 6 may reflect a decrease in sampling resolution due to a slowdown or cessation in shell growth. The relationship between translucent growth lines and $\delta^{18}\text{O}_c$ profiles indicates that these lines were formed during warm seasons (Figures 3, 4). This interpretation is consistent with the fact that the shell margin (i.e., the most recently formed shell material) of the study specimen collected in early August 2004 is not translucent. If this relationship is assumed, the last translucent growth line (Cycle 6) is supposed to have been formed during the warm season of 2003. This result demonstrates that oxygen isotope analysis appears to work well for this species and has the potential to provide useful growth and age estimates.

We analyzed the pattern of the growth curve of shell length against annual growth lines (translucent growth lines) in the specimen based on the number of seasonal cycles of $\delta^{18}\text{O}_c$ (Figure 5). A series of measured data was approximated by the von Bertalanffy equation and a best-fit curve to the data was numerically calculated

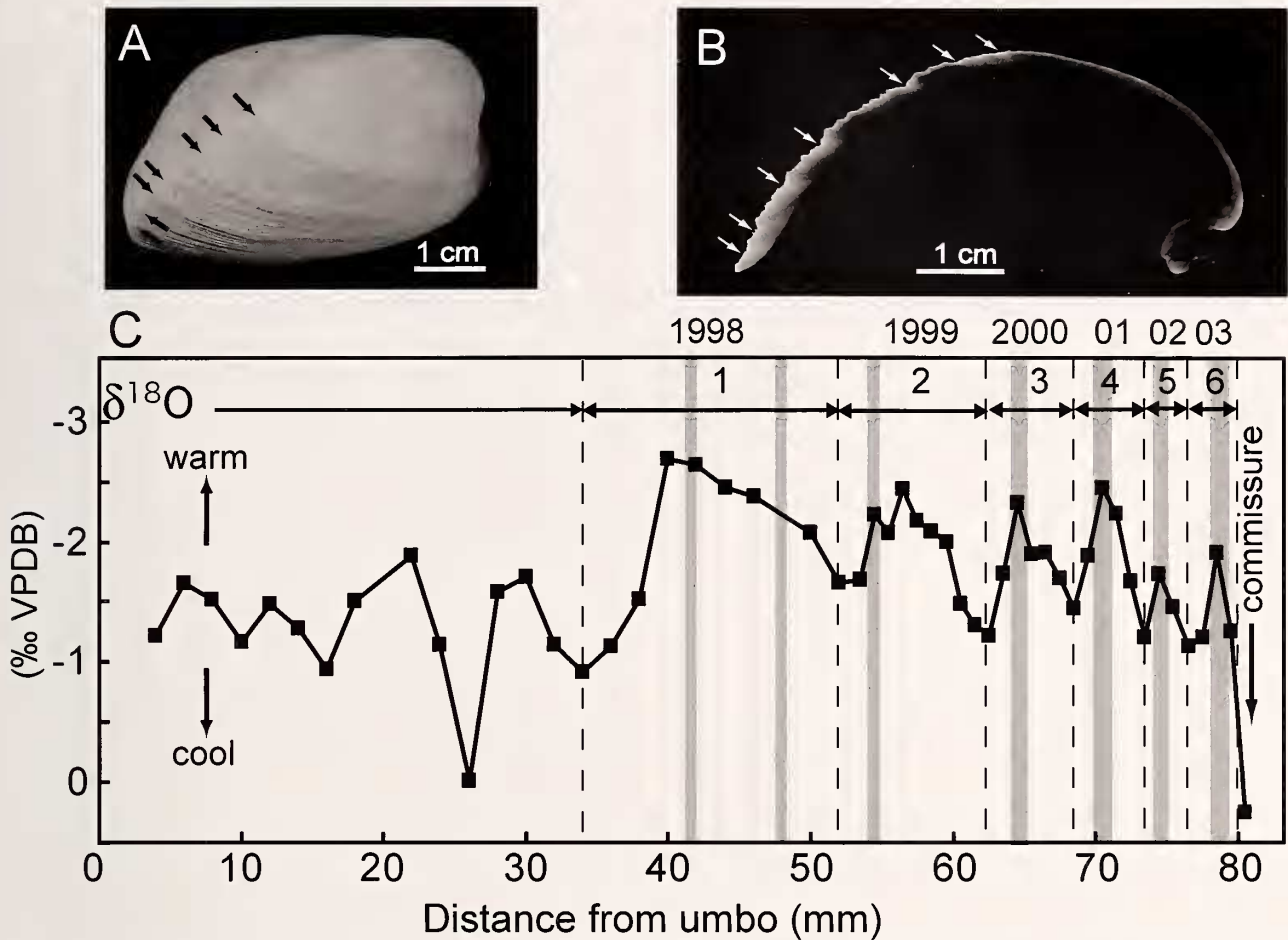


Figure 3. A, Exterior of left valve; B, Optical transmitted-light photograph of the longitudinal shell section. Arrows show translucent growth lines; C, Oxygen isotopic profile for our specimen of *Glossocardia obesa*. Vertical gray bars represent the location of translucent growth lines. Numbers 1–6 are inferred to be cycles bounded by neighboring troughs.

with a least-squares method. Because a seasonal cycle of $\delta^{18}O_c$ was unclear in the early-growth stage, we assumed the following two cases for regression analyses: (1) the bivalve regularly formed each annual growth line in every summer through which the shell has passed (Case I), and (2) the animal failed to form one or a few distinct annual lines in the early-growth stage (Case II). In Case I, the first translucent growth line is supposed to have been formed within 1 yr after hatching. In this case, the beak point with shell length of 0 mm was regarded as the “zeroth” annual increment and the plot of (0,0) was used together with biometric data for regression analysis. On the other hand, if we assume Case II, it took longer than 1 yr until the first annual growth line was formed and the “zeroth” annual increment does not represent the start of the shell growth. In the latter case, therefore, the plot of (0,0) was not used for regression analysis. The fitness of observed data to the von Bertalanffy curve supports

Case II rather than that of Case I (Figure 5). The result suggests that the bivalve started to form annual lines in the second summer or later and lived for at least 8 yr until 2003 (Figure 4).

Combining oxygen isotope analysis with an analysis of absolute growth pattern allow us a reasonable estimation of age and life-history traits of *G. obesa*. Studies on the life-history strategy of this species may provide a clue to reveal the reason why such a “huge” bivalve is a constituent of the refugial community dominated by microshells.

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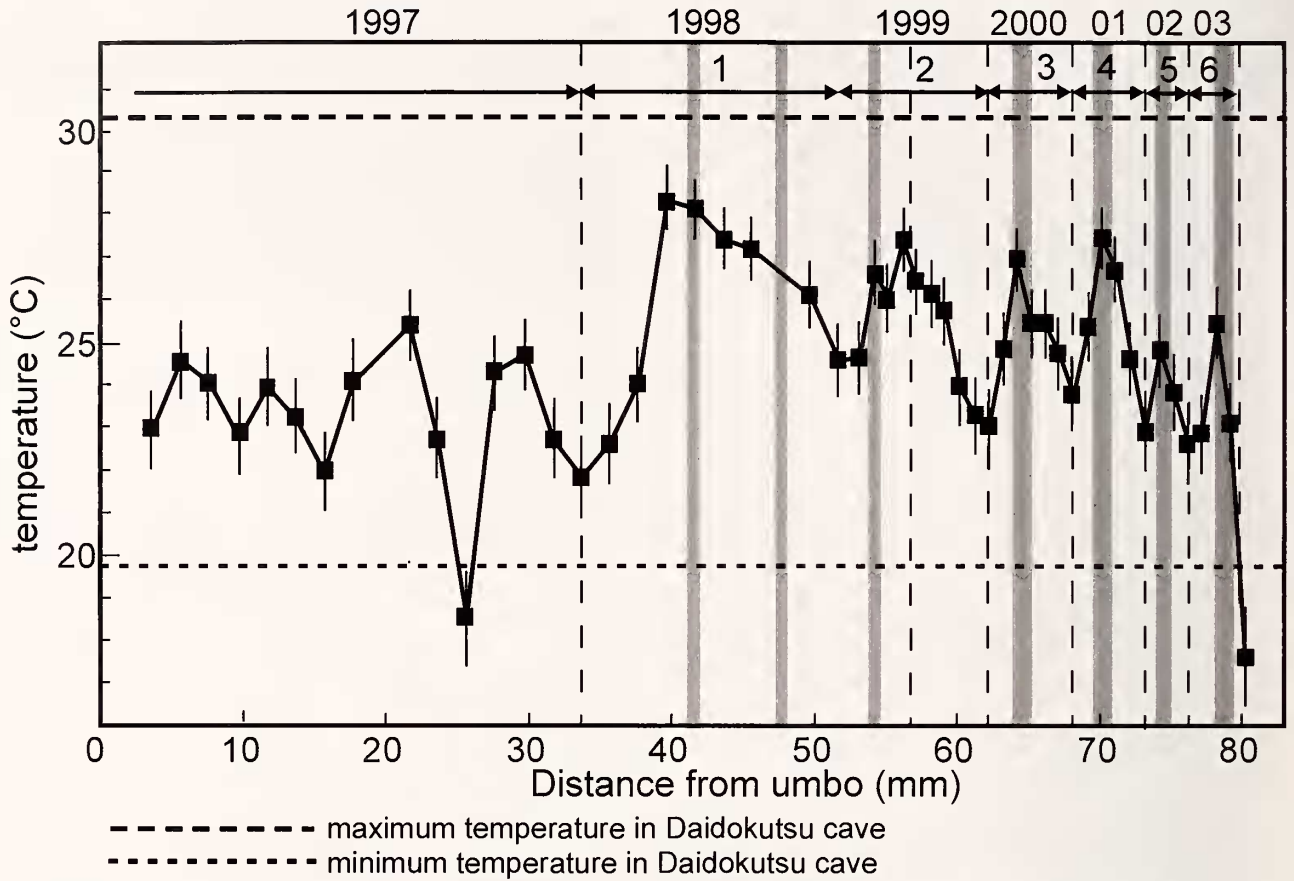


Figure 4. Reconstructed temperature record based on shell oxygen isotope data using the equation of Carré et al. (2005).

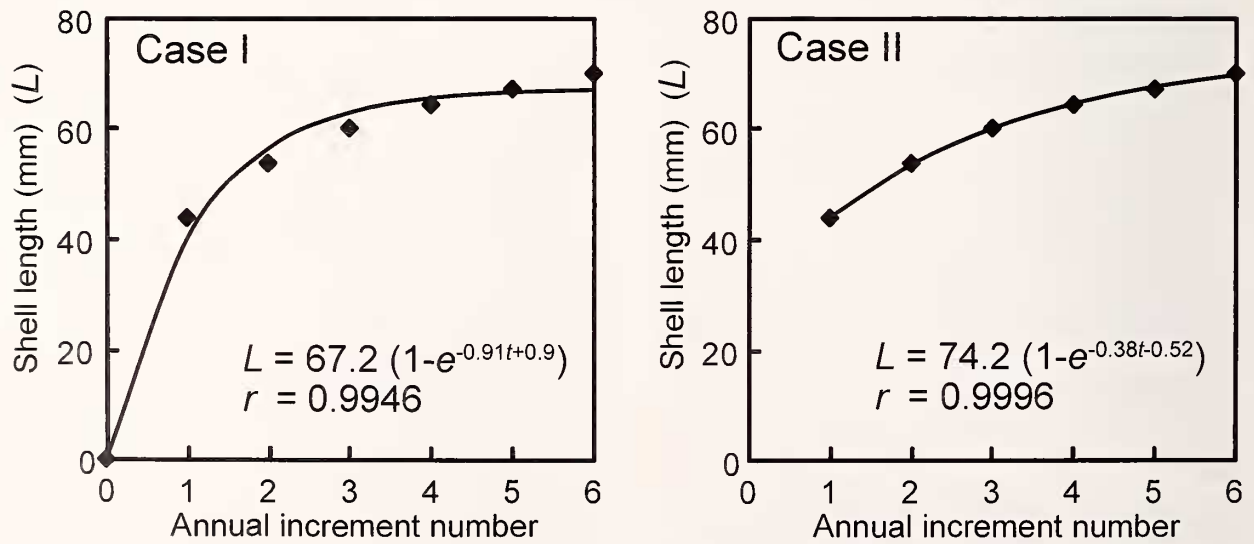


Figure 5. Predicted von Bertalanffy growth curves estimated in the two cases: I, the first annual growth line was formed in the first summer; or II, it was recorded in the second summer or later.

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